

REPLACED BY
ART 34 AMDT

Claims

1. A method for indirectly determining the blood clotting status having the following steps:

5

a) removal of body fluid which contains a protein which can be modified by a vitamin K-dependent γ -carboxylase,

10

b) measuring at least two concentrations selected from a group consisting of a first concentration (C1) of carboxylated protein, a second concentration (C2) of decarboxylated protein and a total concentration (C3) of carboxylated and decarboxylated protein, where the first concentration (C1) is measured using a first antibody (A1), the second concentration is measured using a second antibody (A2) and the third concentration (C3) is measured using a third antibody (A3),

15

c) forming a first ratio (R1) from the first and second concentration (C2)

20

or

25

forming a second ratio (R2) from the third (C3) and first concentration (C1)

or

30

forming a third ratio (R3) from the third (C3) and second concentration (C2),

35

where a concentration (C1, C2, C3) which is necessary for forming the first (R1), second (R2) or third (R3) ratio and is not measured in step b) is calculated in accordance with the following relation:

$$C_3 - C_2 = C_1$$

and

5

d) correlating the first, second or third ratio (R₁, R₂, R₃) with the blood clotting status.

2. The method as claimed in claim 1, where in step b) 10 additionally at least a first competitor (K₁) is used to measure the first concentration (C₁), a second competitor (K₂) is used to measure the second concentration (C₂) or a third competitor (K₃) is used to measure the third concentration 15 (C₃).

3. The method as claimed in claim 1 or 2, where at 20 least one of the antibodies (A₁, A₂, A₃) or at least one of the competitors (K₁, K₂, K₃) is conjugated to a labeling substance, in particular an enzyme, a fluorescent dye, a quencher, a gold particle, a latex particle, a biotin, streptavidin or avidin.

25 4. The method as claimed in any of the preceding claims, where in place of measuring the at least two concentrations as in step b) a combined signal correlating therewith is generated and measured by using two antibodies selected from a group 30 consisting of the first (A₁), the second (A₂) and the third antibody (A₃) and, where appropriate, at least one of the competitors (K₁, K₂, K₃), and is directly correlated with the blood clotting status.

35

5. The method as claimed in claim 4, where the combined signal is a combined color generated in particular by fluorescent dyes, a fluorescent signal elicited by the Förster effect or a

reduction caused by the quencher in a fluorescent signal.

6. The method as claimed in any of the preceding claims, where the body fluid is plasma, blood, saliva, urine or the like.
- 5
7. The method as claimed in any of the preceding claims, where the measurement of the first (C1), second (C2) and/or third concentration (C3) or of the combined signal takes place by an immunological method.
- 10
8. The method as claimed in claim 7, where in the immunological method at least one of the antibodies (A1, A2, A3) is immobilized on a support, in particular a plastic, a magnetic particle, a latex particle, a gold particle, a test strip or a membrane.
- 15
20
9. The method as claimed in any of the preceding claims, where the first (C1), second (C2) and/or third concentration (C3) and/or the combined signal is measured by means of a color reaction or fluorescence detection.
- 25
30
10. The method as claimed in any of the preceding claims, where the protein which can be modified by a vitamin K-dependent γ -carboxylase is prothrombin, factor VII, factor IX, factor X, nephrocalcin or osteocalcin.
- 35
11. A kit for carrying out the method as claimed in any of the preceding claims, comprising at least two antibodies selected from a group consisting of a first antibody (A1) for immunological determination of a first concentration (C1) of the carboxylated form of the protein, a second antibody (A2) for immunological determination of a

second concentration (C2) of the decarboxylated form of the protein and a third antibody (A3) for immunological determination of a total concentration (C3) of carboxylated and
5 decarboxylated protein.

12. A kit as claimed in claim 11, which additionally comprises at least a first competitor (K1) for measuring the first concentration (C1), a second
10 competitor (K2) for measuring the second concentration (C2) or a third competitor (K3) for measuring the third concentration (C3).
13. A kit as claimed in claim 11 or 12, where at least
15 one of the antibodies (A1, A2, A3) or competitors (K1, K2, K3) present is conjugated to a labeling substance, in particular an enzyme, a fluorescent dye, a quencher, a gold particle, a latex particle, biotin, streptavidin or avidin.
20
14. A kit as claimed in any of claims 11 to 13, where the first (A1) and the second antibody (A2) are immobilized on a support, in particular a plastic, a magnetic particle, a latex particle, a gold particle, a test strip or a membrane.
25
15. A kit as claimed in claim 14, where the support is a test strip, and the first (A1) and the second antibody (A2) are each absorbed on a separate field of the test strip.
30
16. A kit as claimed in claim 14 or 15, where a third antibody (A3) conjugated to a labeling substance, in particular an enzyme, a fluorescent dye, a quencher, a gold particle, a latex particle, biotin, streptavidin or avidin is present.
35
17. A kit as claimed in any of claims 11 to 16, where the third antibody (A3) is immobilized on the or

another support, in particular a plastic, a magnetic particle, a latex particle, a gold particle, a test strip or a membrane.

5 18. A kit as claimed in claim 17, where the support is
the or another test strip, and the third antibody
(A3) is absorbed on a field of the test strip.

10 19. A kit as claimed in claim 17 or 18, which
comprises first (A1) and second antibodies (A2) in
each case conjugated to a labeling substance, in
particular an enzyme, a fluorescent dye or a
quencher, where the labeling substances are
selected so that they are able together to
15 generate a combined signal, in particular a
combined color, a fluorescent signal elicited by
the Förster effect or a reduction caused by a
quencher in a fluorescent signal.

20 20. A kit as claimed in any of claims 11 to 19, where
the protein is prothrombin, factor VII, factor IX,
factor X, nephrocalcin or osteocalcin.